Claims 1-14, 16-20, 26-32, 36-68, 73-77, 80-132 and 137-145 stand rejected under 35 U.S.C. §103 as obvious over Cajot *et al.*, Katayose *et al.*, or Srivastava *et al.*, taken with Wills, Liu, Zhang or Brahmwell. The specific grounds for rejection, and applicant's response thereto, are set out in detail below.

II. Personal Interview

Applicant's representative appreciate the Examiner's willingness to permit a personal interview in this case, held on July 27, 1999. At that interview, Applicant's representative discussed the shortcomings of the Cajot et al. reference and presented the Applicant's position (set forth in detail herein) as to why Cajot et al. reference was not believed to be relevant to patentability of the claimed invention. Additionally, the Examiner discussed the concept of "functional p53" and treatment effects. No agreement was reached, but the Examiner indicated that she would consider Applicant's arguments when filed.

III. Rejections Under 35 U.S.C. §112, First Paragraph

Claims 33-35, 69-72, 104-107, 133-136, 140, 141, 144 and 145 stand rejected under 35 U.S.C. §112, first paragraph. The examiner continues to object to claims drawn to combination therapies involving other genes, in addition to p53. The examiner's position is that the specification fails to teach which genes will be effective at augmenting the anti-tumor response of p53, and fails to show that genes will be able to exert their effects.

Applicant respectfully traverses the rejection on the basis that 1) all of the recited genes are recognized to be useful in gene therapy applications, and methods are known in the art for their application, and 2) there is no requirement under §112 that there be shown that the second

gene will "augment" the action of p53. Nevertheless, since the more generic claims, such as claim 1, will cover the specified use of the p53 gene regardless of whether it is used alone or in combination with a second gene, Applicants have canceled the referenced dependent claims in order to progress the case towards allowance.

In addition, the examiner also rejects claims 140, 141, 144 and 145 as being inconsistent with "direct" administration to a tumor. Applicant has canceled these claims as being potentially confusing with respect to the invention now claimed in the independent claims from which they depend. This rejection is therefore now moot.

Reconsideration and withdrawal of both rejections is respectfully requested.

IV. Rejections Under 35 U.S.C. §112, Second Paragraph

Claims 1-14, 16-20, 26-37, 74-77, 80-108, 140 and 144 stand rejected under 35 U.S.C. §112, second paragraph. No grounds for rejection are given for claims 1-14, 16-20, 26-37, 74-77 and 80-108; thus, an effective response is not possible. According to the examiner, claims 140 and 144 are indefinite in reciting intravenous delivery directly to a tumor. Applicant reiterates the remarks set forth in the foregoing section with respect to these claims, which have been canceled. Reconsideration and withdrawal of the rejection is respectfully requested.

V. Rejections Under 35 U.S.C. §103(a)

A. Liu, Wills, Zhang and Brahmwell

Claims 38-68, 73, 109-132 and 137 remain rejected under 35 U.S.C. §103 as obvious over Liu *et al.* (1994) ("Liu") or Wills *et al.* ("Wills") in view of Zhang *et al.* ("Zhang") or Brahmwell. Liu and Willis are cited as teaching *in vivo* delivery and expression of p53 into

tumors using adenovirus. Zhang and Brahmwell are said to teach the benefits of combination therapy. Applicant respectfully traverses.

(i) Tumor Resection Claims (38-68 and 73)

Applicant points out that none of the references relied upon by the Examiner in any way teach or suggest the treatment of microscopic residual tumors. Indeed, the references in no way teach or suggest administering the p53 gene to a tumor bed revealed by resection of all or a portion of the tumor. The Liu reference appears to teach only the administration of the p53 gene in adenovirus to SCCHN cell lines, or directly to the primary tumor grown in skin flaps of nude mice. The only surgery taught by Liu concerns reopening of surgical "flaps" under which the primary tumors are grown. In Liu, the tumor is not removed prior to administration of the adeno-p53 and there is no suggestion that it be removed prior to administration of the adeno-p53.

The same can be said for Wills, which merely teaches the injection of adeno-p53 at the site of growing tumors in nude mice. Wills teaches nothing about combination with surgical therapy and says nothing about the resection of tumor masses prior to gene therapy.

If Applicant has misunderstood or overlooked some relevant teaching in either of these articles, the Examiner is requested to identify the teaching that is being relied upon.

The secondary references also fail to provide a basis for a conclusion of obviousness. The Examiner argues that Zhang suggests treating microscopic residual disease. However, all that Zhang teaches, and all that is apparently relied upon from that reference, is that surgery may at some point in time be used in combination with gene therapy (col. 2, p. 505). Applicant has been unable to identify any teaching relevant to the treatment of microscopic disease. The passage relied upon by the Examiner is so vague that it is virtually irrelevant and could be taken

to mean almost anything or in fact nothing. For example, Zhang's reference to surgery could be taken as suggesting the injection the p53 gene into the primary tumor mass followed by resection of that same tumor mass (as opposed to tumor resection prior to p53 therapy as required by the claim). Alternatively, Zhang could merely be suggesting that surgery is a useful way of actually exposing the tumor itself for injection of the p53 gene. Neither of the foregoing possible interpretations of Zhang would in any way obviate the treatment of microscopic residual tumors after tumor resection.

In fact, when taken in combination with Liu, the logical conclusion is that surgery is to be used merely to reveal an existing tumor mass rather than as a means of treating the primary tumor, followed by gene therapy of any residual microscopic growth. The reason for this is that the only type of "surgery" actually performed by Liu related to simply revealing the tumor followed by adeno-p53 administration directly to the tumor itself (see Liu, page 3663, "After 4 days, the animals were re-anesthetized, and the flaps were re-elevated for delivery [of the adeno-p53 to the tumor].") Thus, the combination of Liu with Zhang would in no way suggest removal of the tumor followed by treatment of the remaining resection bed.

More importantly, though, Zhang must be taken for its face value – and on its face, it says nothing about the treatment of microscopic residual disease. Zhang merely suggests that "someday" it may be possible to treat patients with combination gene therapy and surgery without providing any disclosure of details as to how this might "someday" be carried out.

Lastly, Bramwell is not at all relevant in that Applicant has been unable to identify any suggestion of combination therapy with surgery. If Applicant is in error, the Examiner is requested to point out that passage being relied upon. Barring such an eventuality, Applicant

submits that Bramwell in fact teaches away from combination with surgery, in that it conspicuously fails to suggest such a possibility.

In conclusion, while it may be true that using surgery to enhance the therapeutic benefit of other therapies is known, this is a far cry from teaching or suggesting the specific endeavor of treating, in post-operative fashion, microscopic residual disease that remains after tumor excision. In fact, Zhang does not teach treatment of microscopic residual disease, nor does it even allude to the problem of post-operative reoccurrence. Thus, it is submitted that the rejection is fatally defective in failing to provide a disclosure of an element of the claimed invention.

(ii) Perfusion Claims (109-132 and 137)

With regard to the "continuous perfusion" claims, Applicant respectfully traverses the Examiner's conclusion of obviousness. The Examiner appears to present two separate arguments in support of the rejection. First, the Examiner suggests that the phrase "continuous perfusion" could cover a single bolus injection. Secondly, the Examiner suggests, without support, that contacting the tumor with the virus for extended periods of time would be expected to be advantageous.

In response, Applicant has amended the principal claim, claim 109, to remove the word "continuous" as being confusing, and by adding the requirement for perfusion through the use of a catheter. Support for the use of a catheter can be found in the specification, *e.g.*, at page 33, lines 18 through page 34, line 2, particularly page 33, lines 22-25. This amendment should distinguish the claims from the situation where a single injection is administered intratumorally.

Regarding the Examiner's comment that it was "well known in the art ... that the longer the vector is in contact with the cell, the greater the transduction efficiency," Applicant respectfully requests that such art being relied upon be made of record so that Applicant might respond in a meaningful fashion. If the Examiner is instead relying upon personal knowledge, the Examiner is respectfully requested to appropriately make such personal knowledge of record.

In any event, it is respectfully submitted that the subject claims are now free of the prior art, and reconsideration is requested in light of the foregoing. Indeed, it is incumbent upon the examiner to come forward with specific reasons, based upon the prior art disclosures, as to why those of skill in the art would combine the cited references to arrive at the present invention ***at the time of the invention***. As discussed in the recent case of *In re Dembiczak*, Slip. Op. 98-1498 (April 28, 1999), strict adherence to this methodology will prevent "fall[ing] victim to the insidious effect of a hindsight syndrome wherein that which only the inventor taught is used against its teacher." *Id.* at p. 7, citing *W.L. Gore & Assoc., Inc. v. Garlock, Inc.*, 220 USPQ 303, 313 (Fed. Cir. 1983). To the contrary, the examiner here has viewed ***the present invention*** first, and then assembled odd references which allegedly (though incompletely) disclose elements of the present invention. In so doing, the examiner has committed just the error warned against by *Dembiczak*, *Gore* and a host of other cases, namely, using hindsight as a foundation for the rejection. *Interconnect Planning Corp. v. Feil*, 227 USPQ 543, 547 (Fed. Cir. 1985) ("The invention must be view not with the blueprint drawn by the inventor, but in the state of the art that existed at the time.").

In both of the preceding rejections, the examiner has simply asserted that the claimed invention is obvious. To the extent that the cited art can be said to disclose elements of the present invention, there has been no effort to establish that the references posit their own

combination. However, a long line of cases establishes that the references must provide some basis for their use in conjunction with each other. "Broad conclusory statements regarding the teaching of multiple references, standing alone, are not 'evidence.'" *Dembiczak* at p. 9. Rather, the PTO "must identify specifically ... the reasons one of ordinary skill in the art would have been motivated to select the references and combine them." *In re Rouffet*, 47 USPQ2d, 1453, 1459 (Fed. Cir. 1998).

B. Cajot, Katayose, Srivastava, Wills, Liu, Zhang and Brahmwell

Claims 1-14, 16-20, 26-32, 36-68, 73-77, 80-132 and 137-145 are next rejected under 35 U.S.C. §103 as obvious over Cajot *et al.* ("Cajot"), Katayose *et al.* ("Katayose"), or Srivastava *et al.* ("Srivastava"), taken with Wills, Liu, Zhang or Brahmwell. The primary references are said to teach that provision of p53 to p53-positive tumor cells results in inhibition of tumor cell growth. The secondary references are cited for various aspect of gene therapy. Together, the examiner indicates that these references obviate the treatment of p53-positive tumors *in vivo*. Applicant respectfully traverses.

Ignoring for the moment the issue of whether the secondary references support gene therapy generally, Applicant respectfully submits that the primary references do not, in fact, suggest to one of skill in the art the treatment of p53-positive tumor cells using p53 expression constructs. This is true for at least three reasons. First, as evidenced by the references themselves, there was a considerable difference of opinion in the field as to whether p53-positive cells were susceptible to treatment. Second, there also was some indication that *in vitro* studies, at least with respect to treatment of p53-positive tumors, were not predictive of *in vivo* efficacy. And third, at least one of the studies was fundamentally flawed, from a scientific standpoint,

thereby reducing its probative value to a nullity. In light of these consideration, it is respectfully submitted, as explained in detail below, that no valid *prima facie* case could exist against the rejected claims.

(i) Katayose and Srivastava Evidence Confusion in the Field

As will be explained, Katayose and Srivastava fall far short of an endorsement of treating p53-positive tumors with a p53 expression construct. In fact, a more accurate portrayal of these teachings would be that there was considerable confusion in the field as to whether a p53 positive tumor in an actual human patient (as opposed to *in vitro* or in a nude mouse) could be successfully treated. Indeed, this confusion can be seen as teaching away from treating a p53-positive tumor with additional exogenous p53.

Katayose

Turning first to Katayose, this study employed adeno-p53 constructs to examine the susceptibility of various tumor cell lines (p53 null, p53 mutant, p53 positive) to p53 gene therapy. As abstracted, the results indicated that "tumor cells that were null for p53 prior to infection ... and tumor cells that expressed mutant endogenous p53 protein ... were more sensitive to AdWTp53 cytotoxicity than cells that contained the wild-type p53 ...." This rather non-committal statement is clarified by the last line of the introduction which states that "these studies indicated that an adenovirus vector expressing wild-type p53 is markedly cytotoxic ***to tumor cells that have null or mutant p53 expression*** ..." (emphasis added). No mention is made of p53-positive cells. In addition, the last line of the abstract summarizes the authors conclusions: "These data suggest that endogenous p53 status is a determinant of AdWTp53-mediated cell killing of human tumors." The clear inference is that only p53 null or p53 mutant tumor cells are killed by AdWTp53, not tumor cells that are WTp53.

Katayose indeed actually teaches away from treating p53-positve tumor cells with p53 expression vector. In the Discussion on page 896, first column, second paragraph, it is stated that "There are several possible mechanisms by which high expression of wild-type p53 results in apoptosis in tumor cells devoid of p53 or expressing mutant p53, but not in tumor or normal cells expressing wild-type p53". Thus, Katayose is itself stating quite clearly that expression of wild-type p53 would not be expected to effect apoptosis in a tumor which expresses wild-type p53.

The following additional comments also illuminate what the skilled artisan would take away from Katayose. "As shown in Fig. 3, *A* and *B*, infection of H-358 and MDA-MB-231 [p53 null and mutant, respectively] cells with AdWTp53 completely inhibited cell growth .... In contrast, MCF-7 cells [p53 positive] continued to proliferate although at a slower rate than control cells ...." Page 892, right hand column. "It appears that cells that express wild-type p53 were 5-250 times more resistant to the AdWTp53-mediated inhibitory effect on cell growth when compared with cells expressing no *p53* or mutant *p53*." Page 893, right hand column. "These results indicate that tumor cells null for *p53* or expressing an endogenous mutant *p53* undergo apoptosis following exposure to AdWTp53, whereas tumor cells or normal cells expressing wild-type p53 are resistant to apoptosis." Page 895, right hand column. "... [O]verexpression of wild-type p53 induced programmed cells death (apoptosis) of tumor cells devoid of wild-type p53 or expressing endogenous mutant *p53*, but not in tumor or normal cells expressing wild-type p53." Page 896, left hand column. ***These passages clearly indicate that the Katayose reference cannot be read as providing sufficient motivation for treating p53-positive cells. To the contrary, the reference suggests the opposite, that p53-positive cells are far less susceptible to such treatments.***

Srivastava

Srivastava also provides an insufficient basis for suggesting that one should clinically treat p53-positive cells with a p53 expression construct. In fact, the justification of growth inhibitory effects by a hypothetical mutation in LNCaP p53 is demonstrative of the prior art teaching the opposite of that alleged by the Examiner. While the abstract states that "AdWTp53 vector exhibited a potent inhibitory effect on the growth of all [six] of human metastatic prostate cancer cells ...," the ensuing discussion paints a much muddier picture. For example, the authors state that:

> Since several previous studies did not observe cell growth inhibitory effects of exogenous p53 in tumor cells that already contained endogenous wt p53,[24,25] the inhibitory effects of AdWTp53 on LNCaP cells containing endogenous wt p53 was unexpected. However, in agreement with the previous observations,[18] we also did not detect a growth inhibitory effect of AdWTp53 on breast cancer cells, MCF7 containing endogenous wt p53 (data not shown).

Page 845, right hand column.

However, a later statement makes it clear that Srivastava believes that there must be another p53 function that is being supplied by the exogenous wt p53 in these cells: "However, it is possible that some as yet unknown function of p53 is defective in LNCaP cells..." see page 847, second column. This suggests that one skilled in the art was directed away from the idea of clinically treating p53 positive cells with a p53 expression construct. Otherwise, the results in LNCaP cells would not need the justification afforded by this statement. Thus, reading Srivastava, one of skill in the art would not be led to treat p53-positive cells with p53 gene therapy with any likelihood of success.

Taken together, Srivastava and Katayose present a very unclear view of whether the p53 status of a tumor cell is important in determining whether or not p53 gene therapy will be successful. In fact, a more likely interpretation, based on the data, is that p53-positive tumors are

much less likely to respond to such therapy, and if they do respond, they do so far less than do other tumors.

(ii) Extrapolating from In Vitro to In Vivo is Problematic at Best

According to the examiner, it would have been obvious, looking at the alleged p53-positive cell inhibition "successes" of Katayose, Srivastava and Cajot, to then move into the clinical realm according to the teachings of the secondary references. However, as the PTO is quick to tell applicants, extrapolation from *in vitro* data to *in vivo* efficacy is, at best, problematic. That position is, apparently, even true in this particular instance.

In Srivastava, it is noted that "[a] recent study has described an intriguing result in which an adenovirus-p53 expression vector did not inhibit the *in vitro* growth of a metastatic variant of LNCaP cells; however, the growth of these cells was inhibited *in vivo*.[31] This is evidence that there is at least some level of difficulty in predicting, at least in the context of treating p53-positive cells, whether *in vitro* results will hold in an *in vivo* environment. Additionally, the *in vitro* results referenced were contradictory to the data presented in Srivastava leaving doubt as to the success of *in vivo* p53 treatment of LNCap cells. Further evidence for the lack of a reasonable expectation of success is the fact that the investigators did in fact not report the treatment of LNCaP tumors in an *in vivo* mouse model, as implied in the final sentence on page 847 second column. As such, it is apparent that the examiner's straightforward extrapolation not only is not merited, but it is not supported by the evidence of record.

(iii) Srivastava and Katayose Are Not Prior Art

Applicant would further point out that neither Srivastava nor Katayose are prior art to the present application. The present application is entitled to a filing date of November, 1995 – this

date has already been implicitly recognized by the Examiner as an appropriate priority date for the subject claims in that the Clayman and Liu I references were successfully removed through the earlier filed Clayman Rule 131 declaration. Thus, neither of these two references are available under 102(b)/103. The Clayman *et al.*, January, 1995 reference (*Cancer Research*, vol. 55, pp. 1-6) was successfully removed through the filing of the Clayman Rule 131 declaration. Thus, this should sufficiently demonstrate on the record that the invention of the subject claims was made in this country prior to the publication dates of the Srivastava and Katayose references. If the previously filed Clayman Rule 131 declaration is in any way insufficient to remove the Srivastava and Katayose references, Applicant would be pleased to provide any additional declarations of Dr. Clayman as required by the Examiner.

(iv) The Prior Art Teaches Away/Failure of Others

There are a number of additional references that must be considered as teaching away from a conclusion of obviousness, and indeed further evidence the confusion in this field. These include the references of Baker *et al.* (*Science*, 249:912, 1990) and Casey *et al.* (*Oncogene*, 6:1791, 1991) (copies enclosed). Each of these references demonstrates those *in vitro* attempts to achieve suppression of the neoplastic phenotype in p53-positive tumors failed to demonstrate suppression.

The examiner must consider the entirety of the prior art when assessing the obviousness issue, including evidence of failure of others and teaching away. The Baker and Casey references demonstrate failure in *in vitro* studies using p53 to suppress the growth of p53-positive cells. Such teachings must be taken as teaching away from pursuing the clinical application of p53 therapy in humans having wt p53 tumors. Furthermore, when these studies

are further taken into consideration with the observation that *in vitro* results with p53 do not necessarily correlate with *in vivo* observations (see section (ii) above), the only reasonable conclusion is that there is no predictable success in the clinical application of p53 gene for the treatment of tumors expressing wild type p53.

(v) The Cajot Studies are Fatally Flawed

Despite what Cajot may or may not say, Applicant submits that the previously discussed confusion in the art regarding p53-positive cells and their treatment with p53 gene therapy is more than sufficient to rob the examiner's alleged *prima facie* case of the requisite likelihood of success. However, the Cajot paper suffers from an additional defect that should be noted on the record. It is submitted, respectfully, that this defect precludes reliance on Cajot for the simple reason that its results are invalid.

In order to understand more fully the problems with Cajot, it is first necessary to explain their studies in some detail. Human lung cancer cells were transfected with vectors containing either wild-type or mutant p53 under the control of the CMV promoter and the neomycin resistance gene under the control of the SV40 promoter. Cells were subjected to geneticin selection for 3-4 weeks. Thus, stable transformants were selected. The authors concluded that, because few colonies were observed with wild-type p53 expressing clones, there was an inhibitory effect on tumor cell growth by the p53 expressed in these cells.

What Cajot did not account for was the potent down-regulation of the SV40 promoter by p53. Subler *et al.* (1992); Jackson *et al.* (1993); Perrem *et al.* (1995) (attached). Of particular interest is the Subler *et al.* report, which also indicates the a mutation in p53 at position 143, which is the same as Cajot mutant p53, did not show this inhibitory effect.

Looking at the Cajot data, what does this all mean? Well, for one, the difference in the transfection efficiencies seen between wild-type and mutant p53 can be explained by the fact that wild-type p53 shuts down the SV40, eliminating neomycin expression, and thereby eliminating cells because of geneticin toxicity, ***not because of tumor inhibition***. The skilled artisan, being aware of both Subler *et al.* and Cajot, would therefore have dismissed Cajot's observations as invalid.

Even Cajot *et al.* bears out the notion that the remaining tumor clones survived because the p53 gene being expressed became mutated, thereby avoiding the SV40 down-regulation of neomycin: "In contrast, no normal-size transcript characteristic of exogenous p53 was detected in any of the wild-type p53 clones analyzed." Page 6957, right hand column. Further, they state that "[n]o increase in the expression level of this M_r 53,000 band was observed in the wild-type p53 clones, which correlates with the absence of normal-size exogenous transcript detected by Northern analysis." Page 6957, right hand column. Also, in Fig. 3, it should be noted that the tumorigenicity studies were conducted with a clone that admittedly does not express a wild-type p53 product, as the transcript is larger than normal (1.8 kB), and the protein is smaller (45 kD), than true wild-type p53. Thus, based on a singular experiment using an admittedly mutant p53, there is no valid conclusion that can be drawn from the study regarding the effect of ***wild-type*** p53 on tumorigenicity in p53-positive tumor cells.

(vi) Current Clinical Data Supports the Present Claims

The current clinical data supports a conclusion of surprising and unexpected results in the context of the clinical application of the present invention. The clinical data further provides support for the conclusion that when applied in a clinical context, the invention is applicable in

the treatment of wt p53 expressing tumors virtually to the same degree as in the context of non-wt p53 expressing tumors. This is most surely a surprising and unexpected result.

In the Phase I study reported in Clayman *et al., J. Clin. Oncol.* 16:221-2232 (1998) (enclosed), thirty-three patients with recurrent head & neck cancer were treated with intratumoral injections of Ad-p53. The treatment regimen consisted of at least one course of Ad-p53 (three times a week for two weeks). Of these, eighteen patients had non-resectable tumors (and received multiple treatment courses with two week rest between courses; continuing until disease progression or withdrawal of consent), permitting post-treatment assessment of tumor progression. Of these eighteen, twelve were p53+ by sequencing of tumor cell DNA. Of these twelve, two patients had greater than 50% tumor regression, four had stable disease, five had progressive disease, and in one the outcome of treatment could not be evaluated. By comparison, of the remaining six non-resectable patients, one was non-evaluable for p53 status. Of the five patients with mutated p53 genes, four exhibited progressive disease while two exhibited stable disease. Of the 33 patients entered in the study, the remaining 15 underwent complete resection of their tumor three days after a single course of treatment, and could not be rigorously assessed for clinical response.

Similarly, phase II Clinical data has recently become available for the treatment of head & neck cancer using Ad-p53. As discussed in the accompanying declaration of Dr. James A. Merritt, tumors of various p53 status (as determined by sequencing of exons 1-10) have been treated with Ad-p53. These Phase I and II clinical data demonstrate that p53+ tumors were susceptible to treatment by Ad-p53. A summary of the data are show below:

SUMMARY OF PHASE II CLINICAL DATA BY p53 STATUS

p53 status*	CR	PR	SD	PD
p53+	0	2	6	21
p53-	0	1	6	27

* - as determined by sequencing of exons 1-10
CR – complete response; PR – partial response; SD – stable disease; PD – progressive disease

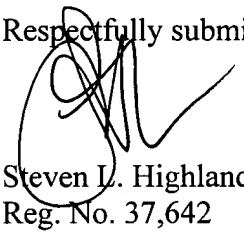
Scoring a complete response as 3, a partial response as 2, stable disease as 1, and progressive disease as 0, patients with p53-positive tumors had an efficacy rating of 0.42, as compared to 0.29 for those having p53- tumors. Using this analysis, Ad-p53 was even ***more*** effective at treating p53+ tumors than p53- tumors.

This result could not have been predicted from the cited art. Thus, it again is respectfully submitted that those of skill in the art would not have found the present invention obvious.

VI. Summary

In light of the foregoing amendments and remarks, applicant submits that all claims are in condition for allowance and solicit an early indication to this effect. Should Examiner Hauda have any questions regarding this response, she is invited to contact the undersigned at the telephone number listed below.

Respectfully submitted,



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